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Concurrent extrahepatic autoimmunity in Autoimmune Hepatitis: Implications for diagnosis, clinical course and long term outcome

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Short title: Extrahepatic autoimmunity in AIH

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Abbreviations: AIH, autoimmune hepatitis; AITD, autoimmune thyroid disease; ANA, antinuclear antibody; CEHAID, concurrent extrahepatic autoimmune disease; HCC, hepatocellular carcinoma; IAIHG, International Autoimmune Hepatitis Group; LKM, liver-kidney-microsomal antibody; LT, liver transplantation; SMA, smooth muscle antibody; UC, ulcerative colitis.

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Abstract

Background: Concurrent extrahepatic autoimmune disease (CEHAID) associated with autoimmune hepatitis (AIH) have been incorporated into the diagnostic criteria stipulated by the International Autoimmune Hepatitis Group (IAIHG). Large comprehensive cohort data on the extrahepatic autoimmunity in AIH remain scanty. **Aim:** To systematically assess features and clinical impact of CEHAID on AIH. **Methods:** Clinical records of 562 patients with AIH from two tertiary centres in the United Kingdom were retrospectively reviewed. **Results:** Prevalence of CEHAID in patients with AIH were 42%. Autoimmune thyroid disease was the commonest CEHAID associated with AIH (101/562, 18%). Autoimmune

skin diseases were more prevalent in AIH-2 than AIH-1 (21.9% vs.7%, p=0.009). Personal history of CEHAID was more commonly found in AIH patients with than without first degree family history of CEHAID [(48/86, 55.8% vs 169/446, 37.9%), p=0.002]. AIH patients with CEHAID were more often female [201/236 (85.2%), p=0.008], had higher post-treatment IAIHG score (22 vs. 20, p<0.001), less reactivity to smooth muscle antibodies (49.8% vs 65%, p<0.001), more likely to have mild fibrosis at diagnosis (20.9% vs. 6.5%, p<0.001), less often had ascites (6.3% vs. 13.6%, p=0.008) and coagulopathy (1.18 vs. 1.27, p=0.013) at presentation. Presence of CEHAID, however, did not significantly affect disease progression, prognosis and survival in AIH. **Conclusions:** Our study confirms the strong association of CEHAID with AIH. Association between personal and familial extrahepatic autoimmunity especially among first degree relatives was evident. Presence of CEHAID may influence clinical phenotype of AIH at presentation but without notable impact on the long term clinical outcomes.

Key words: Autoimmune thyroiditis, autoimmune skin disease, extrahepatic autoimmune diseases, family history, first degree relatives

Key Points

- This is the largest cohort study to systematically assess patterns and clinical impact of CEHAID in AIH.
- AIH is strongly associated with CEHAID. Autoimmune skin diseases were more prevalent in AIH-2 than AIH-1.
- This study reinforces the close association of extrahepatic autoimmunity among AIH patients and their family particularly first degree relatives.

- This is the first study to observe that presence of CEHAID may influence the reactivity of the circulating smooth muscle antibodies and effect the clinical phenotype of AIH at index presentation, albeit, without seeming to impact on disease progression and clinical outcomes.

Introduction

Autoimmune hepaitits (AIH) is an immune-mediated liver disease which can present in acute or chronic forms, and may lead to cirrhosis and liver failure if untreated. It is characterised by elevated transaminases, raised Immunoglobulin G levels, histological features of interface hepatitis with a lymphoplasmacytic infiltration and the presence of autoantibodies in serum (1–3).

The diagnosis and management of AIH could be challenging as AIH represents a chameleon disease with protean clinical manifestation and significant heterogeneity in relation to clinical course and outcome. Indeed, the clinical features are not confined to the liver. Concurrent extrahepatic autoimmune disorders (CEHAID) including autoimmune thyroiditis, connective tissue disease and inflammatory bowel disease are frequently associated with AIH. CEHAID may predate, coincide or even occur years after the diagnosis of AIH. This association has been recognised and incorporated into the original and revised International Autoimmune Hepatitis Group (IAIHG) scoring systems as an aid to codifing the diagnosis (4,5). The frequency of CEHAID in AIH has been derived predominantly from case reports (6–12) and a few cohort studies (13–15). Large cohort studies to systematically assess the features and effect of CEHAID in AIH are lacking.

On the other aspect, since AIH arises in genetically susceptible individuals, there may be a close relationship between personal and familial hepatic and extrahepatic autoimmunity. Although up to 43% of AIH patients have been reported to have a family history of CEHAID

in their first degree relatives, commonly with thyroid disease and type 1 diabetes (16,17), the association of extrahepatic autoimmunity among AIH patients and their family especially first degree relatives remains unexplored.

In this study, we sought to describe the patterns of CEHAID, the association of personal and familial extrahepatic autoimmunity in AIH and finally to evaluate the impact of CEHAID, if any, on the clinical course and long term outcomes.

Patients and Methods

Two well-established and updated databases of patients with AIH attending the Institute of Liver studies at King's College Hospital, London between 1971 and October 2015, and the Department of Hepatology, Brighton and Sussex University Hospital, Brighton between January 2005 and June 2014 were reviewed. A total of 562 consecutive patients with AIH were identified [455 definite AIH, median post treatment IAIHG score 21 (18-28) and 107 probable AIH, median post treatment score 16 (11-17)]. The diagnosis of AIH was made based on the revised IAIHG scoring system (5). Median duration of follow-up was 123 months (0.5-544). Patients' records were systematically reviewed and examined with regards to the frequency and patterns of CEHAID as well as the association between personal and family history of CEHAID among AIH patients and their first and second degree relatives. Finally, two groups of AIH patients with and without CEHAID were compared in relation to the clinical, laboratory and histological features, response to therapy, clinical outcomes and survival.

As described in our previous studies, standard diagnostic criteria for the presence of AIH was fulfilled (18–20). There was no corroborative history of concomitant use of hepatotoxic drugs except for eleven patients who had diagnosis of drug-induced AIH (20).

Liver biopsy was available at diagnosis in 471(83.8%) of 562 patients with AIH. Histological assessment of the severity of liver inflammation and the degree of fibrosis was based on the scoring system proposed by Batts and Ludwig (21). In this system, necroinflammatory activity is graded as 0 = none, 1= minimal, 2 = mild, 3 = moderate and 4 = marked activity +/- bridging collapse or multiacinar necrosis, whereas the degree of fibrosis is staged as 0 = none, 1 = portal, 2 = periportal, 3 = septal/bridging fibrosis including incomplete cirrhosis and 4 = cirrhosis.

AIH-1 was defined by the presence of antinuclear antibody (ANA) or smooth muscle antibody (SMA) or both. AIH-2 and autoantibody negative AIH were classified by the presence of anti-liver-kidney microsomal-1 (anti-LKM1) and absence of all the autoantibodies described respectively.

Associated diagnosis of CEHAID and family history of autoimmune diseases were searched and retrieved via the the hospital electronic records, clinical letters and medical casenotes. All these diseases have been diagnosed and confirmed based on the international criteria, when available.

Time to diagnosis was defined as the time from the first onset of symptoms or first detection of liver dysfunction to the formal diagnosis of AIH. The mode of presentation refered to the acuity of the initial illness or symtomatology, and was defined as 'acute' if symptom onset to diagnosis was $\leq$ 6 months, 'insidious'> 6 months and 'asymptomatic' if the patients had no obvious signs or symptoms of liver disease and the diagnosis of AIH was first discovered based on the incidental finding of abnormal liver tests either during routine health screening or during evaluation of a non-hepatic illness. Follow-up duration was defined as the time of the diagnosis was first made until the last outpatient appointment in the clinic, or death or liver transplantation (LT) (18,19).

All patients with AIH were treated according to standardised protocols published previously (18,19,22). Response to treatment and relapse were defined in accordance with the revised criteria of the IAIHG (5), and partial or no response to initial therapy according to the original criteria (4).

Disease progression was defined as the development of cirrhosis in non-cirrhotic patients based on imaging and/or histology during follow-up, worsening of the fibrosis scores on repeat histology, when available, despite on optimum immuosuppression, and the occurrence of clinically significant liver-related complications including episodes of decompensation, development of hepatocellular carcinoma (HCC), death or LT in cirrhotic patients.

This study was approved by the Research Ethics Committee of King's College Hospital (04/Q0703/23) and is part of an ongoing research project in AIH for which patients have given informed consent.

Statistical analysis

Results were analysed using SPSS version 21.0 (IBM Corp, Armonk NY). Continuous variables were expressed as median (range). Categorical variables are expressed as actual numbers and percentages. Analysis of variance was used to compare the differences in variable between the two groups. Group comparisons of categorical variables were analysed with $\chi 2$-test or Fisher's exact test if the expected cell frequency is less than 5. The Mann–Whitney test was used for the evaluation of continuous variables.

End-points and censoring date for survival analysis were taken as the time of most recent clinic visit or date of death or LT. Death from complications of cirrhosis, progressive liver failure and HCC were considered to be liver-related. Survival rates between 2 groups of

AIH patients with and without CEHAID were calculated according to the Kaplan-Meier method and compared using the log-rank test. Univariate analysis using Cox's proportional hazards regression model for survival analysis was adopted to assess the predictive value of a number of variables on survival outcome. This was followed by multivariate analysis to identify independent predictors of adverse outcome. A value of $P < 0.05$ was considered to be statistically significant. Missing data were <10% in all categories (except where specified, i.e. histology and variables for clinical outcomes) and were excluded on a per-analysis basis.

Results

Frequency of CEHAID in AIH

Two hundred and thirty-six of 562 AIH patients (42%) had at least one associated CEHAID. Among these, 168 (29.9%) had one associated CEHAID, 53 (9.4%) had two, 10 (1.8%) had three, 3 (0.5%) had four, and one each had five and seven associated CEHAIDs respectively (Fig. 1).

In total, 58 different diagnoses of CEHAID were identified. Of these, 26/253(10.2%), 89/253(35.2%) and 138/253(54.6%) were diagnosed at presentation, during the follow-up and preceding the diagnosis of AIH, respectively. The chronological diagnosis of 75 CEHAIDs in relation to the diagnosis of AIH were uncertain. The frequency of associated CEHAID in AIH was depicted in Table 1. Other rarer CEHAIDs are not shown.

Family History of concurrent hepatic and CEHAID in AIH

A positive family history of CEHAID was identified in 92/532 (17.3%) patients with AIH, and of these, 51/92 (55.4%) had concurrent extrahepatic autoimmunity in their personal history. This was significantly more prevalent when compared to patients without family history of CEHAID, 166/440 (37.7%) (p=0.002). Eighty-six patients (86/532, 16.2%) with a

positive family history had at least one first degree relative with an associated CEHAID. Amongst these, 57/86 (66.2%) had one first degree relative with CEHAID, 22/86 (25.6%) had two, 4 (4.7%) had three, 2 (2.3%) had four and one (1.2%) had six first degree relatives with CEHAID. In the same context, personal history of CEHAID was more commonly found in AIH patients with than without first degree family history of CEHAID [(48/86, 55.8% vs 169/446, 37.9%), p=0.002]. A postive family history of CEHAID amongst second degree relatives was also present in 14 AIH patients (2.6%) but there was no significant difference between patients with and without extrahepatic autoimmunity in their personal history [9/14 (64.3%) vs. 208/518 (40.2%), p=0.070].

The most common CEHAID among family members was thyroid disease (45/92, 48.9%) followed by diabetes mellitus (26/92, 28.2%), connective tissue disorders (21/92, 22.8%) and autoimmune skin disease (8/92, 8.7%). The prevalence of a family history of an autoimmune liver disease in AIH cohort was 7/532 (1.3%). Family history of AIH and PBC was found in 4 patients (0.75%) and 3 patients (0.56%), respectively.

Clinical patterns of CEHAID in AIH

Autoimmune thyroid disease (AITD) was the commonest CEHAID in AIH patients (18%), followed by connective tissue disorders (12.3%) and autoimmune skin diseases (8%) (Table 1). Hypothyroidism was more prevalent than hyperthyroidism [79/101 (78.2%) vs 22/101 (21.8%), p<0.001]. Based on the serological subtypes of AIH (Table 2), AITD were more common in AIH-2 and autoantibody negative AIH than AIH-1 (31.1% vs 15.9% and 32% vs 15.9% respectively, p<0.05). Hyperthyroidism was more prevalent in AIH-2 (12.5 vs 3.1%, p=0.024) whereas hypothyroidism was more common in autoantibody negative AIH than AIH-1 (25% vs 12.9%, p=0.014). Interestingly, autoimmune skin lesions were more frequently diagnosed in AIH-2 than AIH-1 (21.9% vs 7%, p=0.009). The main skin lesions in AIH-2 included vitiligo, leucocytoclastic vasculitis, urticaria and alopecia areata.

Impact of CEHAID on the clinical course and outcome of AIH

Tables 3 and 4 illustrate the demographics, clinical presentation, biochemical and histological aspects of AIH patients with and without CEHAID. Women were more likely to have CEHAID than men (201/248, 81% vs 35/79, 44.8%, p=0.008). Interestingly, AIH patients with CEHAID had lower occurence of ascites and had less prolonged protrombin time/International Normalised ratio (INR) at presentation (6.3% vs. 13.6%, p=0.008 and 1.18 vs. 1.27, p=0.013), respectively. Moreover, AIH patients with CEHAID frequently had mild fibrosis (stage 1) on histology at diagnosis when compared to patients without CEHAID [(28/134 (20.9%) vs. 13/200 (6.5%), p<0.001]. There was, however, no difference in terms of severity of the histological inflammation at diagnosis between the 2 groups. The incidence of cirrhosis at presentation (based on histology and /or imaging studies) did not differ significantly between AIH patients with and without CEHAID (31% vs. 35.6% p=0.261). The coexistence of associated CEHAID was also independent of the modes of presentation.

Patients with CEHAID tended to have joint pain as their index presentation compared to their counterparts without CEHAID [18/221 (8.1%) vs. 9/295 (3.1%), p=0.01]. The frequency of ANA positivity did not differ significantly in the presence of CEHAID. Nonetheless, the median peak ANA titre was significantly higher in patients with CEHAID (1:80 vs 1:40, p <0.001). In contrast, SMA positivity was more prevalent in AIH patients without CEHAID (65.1% vs 49.8%, p<0.001). Further analysis revealed a higher proportion of patients without CEHAID had a median SMA titre of >1:80 (41.2% vs 30%, p=0.009) and significantly lower proportion with a median SMA titre of <1:40 (41.8% vs 53.8%, p=0.006) when compared to patients with CEHAID.

There were, however, no significant differences between AIH patients with and without CEHAID pertinent to treatment modality utilised, treatment responses, number of relapses and long term clinical outcomes including disease progression, development of HCC and episodes of decompensation leading to LT and liver-related death during the follow-up period (Table 5). There was also no significant difference in survival between AIH patients with and without CEHAID ($p = 0.563$ by log-rank comparison) (Figure 2). Although univariate analysis revealed ascites and INR as one of the variables associated with reduced survival, serum albumin at presentation appeared to be an independent predictors of reduced survival following multivariate anlaysis (Table 6).

Discussions

This is hitherto the largest cohort study to systematically interogate the patterns and clinical impact of CEHAID in patients with AIH. Akin to cohort studies from Italy and Germany (14,15), we recorded a high frequency of CEHAID in AIH (42%), with AITD being the commonest (18%). Additionally, we found that hypothyroidism was more commonly diagnosed than hyperthyrodism and this seemed to be influenced by the type of AIH. Another interesting finding was that autoimmune skin diseases were more frequently associated with AIH-2 than AIH-1. However, this association was not evident in a South American study (13). On the contrary, the prevalence of skin diseases in our study was comparable to the report by Homberg *et al.* describing a cohort of LKM positive AIH patients of North European ancestry (21.9% vs. 27.2%)(23). Coeliac disease was found in 1.4% of our AIH pateints. This finding accords with the frequency reported in a German study (1.1%) (15) but slightly lower than the reports from the Italian and Dutch groups (3.5% and 2.8%, respectively) (14,24).

Interestingly, our data showed that AIH patients with CEHAID tended to present less often with ascites and coagulopathy at presentation compared to patients without CEHAID. In addition, they were more often had mild fibrosis on histology at diagnosis. Our study is the first to observe this clinical association. There may be two possible hypotheses. Firstly, the presence of CEHAID could have led to an early diagnosis of AIH and rendered a "protective" effect. This was supported by the observation that more than half of the CEHAID predated AIH diagnosis and there was no significant difference pertinent to the histological severity and the clinical course or outcome between the two groups of patients. However, the time from the first onset of symptoms or first detection of liver dysfunction to the formal diagnosis of AIH was similar irrespective of presence of CEHAID. Due to the retrospective nature of this study, whether or not the respective subspecialists would monitor liver functions during follow-up with CEHAID or suspect AIH on first abnormal liver dysfunction and refer to hepatologist remains a potential limiting factor which may lead to inaccurate estimate of the time to diagnosis of AIH.

Secondly, AIH patients with CEHAID, on the contrary, could have a more aggressive phenotype as they were more likely to have advanced fibrosis (stage 3 and 4) at diagnosis compared to AIH patients wthout CEHAID (158/200, 79% vs. 81/134, 60.4%, $p<0.001$) (Table 4). Nevertheless, there was no difference between fibrosis stage 3 and 4 patients with or without CEHAID in terms of death/LT (19/66, 28.2% vs. 45/137, 32.8%, $p=0.560$), HCC development (4/66, 6.1% vs. 7/137, 5.1%, $p=0.751$), disease progression (30/63, 47.6% vs. 54/142, 38%, $p=0.198$) and overall survival ($p=0.575$ by log rank comparison) (data not shown). Furthermore, Cox-regression model for survival analysis using univariate and multivariate analysis demonstrated that neither presence of cirrhosis, ascites nor prolonged INR at diagnosis as the independent predictor for reduced survival (Table 6). Therefore,

though the clinical phenotype for AIH patients with CEHAID appears aggressive at presentation, it does not seem to portend poor clinical outcomes.

In a recent study by Muratori *et al*., more altered liver biochemistry i.e higher serum bilirubin and tramsaminitis, was observed in AIH patients without CEHAID, but no statistical significance was attained (14). It is worth noting that there was no documentation of coagulation profile, occurrence of ascites and fibrosis stage in their report. They noted that AIH patients with CEHAID tended to be asymptomatic at presentation and they attributed this to a more closer monitoring in this group of patients for other autoimmune diseases, therefore any liver related complications could be detected earlier and associated with ascertainment bias. This could provide a possible account for our findings but we did not observe more asymtomatic patients associated with CEHAID in our cohort.

Whether or not the presence of CEHAID could be a "protective" mechanism to a more aggressive clinical phenotype in AIH at presentation remains to be elucidated. Nonetheless, this clinical observation may shed light on the intriguing interplay between the shared genetic predisposition of CEHAID and the clinical manifestation of AIH at initial presentation.

The other interesting finding in our study is that SMA positive AIH patients had a lower prevalence of CEHAID than SMA negative patients. It is noteworthy that in another UK population study with PBC, AMA positive PBC patients were also reported to have a significantly lower prevalence of an additional autoimmunity than AMA negative patients (25). SMA reactivity in titre >1:80 was reported to be associated with evidence of histological disease activity before treatment in a study of 117 patients with AIH (26). However, in our study, the higher prevalence of SMA reactivity in a titre of >1:80 in AIH patients without CEHAID did not translate to a more severe histological inflammation when compared to patients with CEHAID.

Joint pain has been well-documented as an extrahepatic symptom in 10-60% of AIH patients at index presentation (1,16,27,28). Arthralgia was reported in 5.2% of our AIH patients. It was the only symptom that was more common in isolation in AIH patients with CEHAID. This is potentially contributed by the presenting symptoms of the concomittant rheumatological diseases as one of the associated CEHAID. In this study, we also demonstrated that CEHAID tended to cluster in female patients. Except for a South American study in which they found no gender predilection for the association of CEHAID (13), our finding was in congruence with previous studies (14,16,29,30). The difference may be accounted for by the modulation of immune systems by both the hypothalamic-pituitary-gonadal system and sex hormones whereby androgens and oestrogens suppress and enhance immunity, respectively (31).

It is well-recognised that patients with autoimmune disease are prone to having additional autoimmune conditions affecting different organs, which may coexist in individual patients and their families (25,32). Indeed, the coexistence of concomittant personal or family autoimmunity confers a score of +1 and +2 in the the original and revised IAIHG scoring systems respectively (4,5). In our study, 16.2% of AIH patients had family history of CEHAID in first degree relatives, which was in parallel with our previous report with 234 AIH patients (19.6%) (18), but relatively lower when compared to other studies by Van Gerven *et al.* and Gregory *et al.* (42% and 43% respectively) (16,17). However, more importantly, we reported that a significantly higher proportion of AIH patients who had a positive family history of CEHAID, had concurrent extrahepatic autoimmunity in their personal history when compared to the counterparts without family history of CEHAID. This association was significantly more evident in first degree relatives than second degree relatives. However, the results may be limited by the retrospective nature of this study whereby the detail documentation of the personal and familial history of CEHAID depends

on individual physicians' discretion. This may potentially lead to underestimate of the real frequency in this cohort. Nevertheless, to our knowledge, this is the first study to demonstrate the close link between personal and familial extrahepatic autoimmunity in an AIH cohort and emphasize the pivotal role of personal and family history of CEHAID in the IAIHG scoring system.

Conclusions

In this study, we have confirmed strong association of extrahepatic autoimmunity in patients with AIH. Recognition of CEHAID is important since it may reflect specific disease phenotypes and serves as a clue for subsequent diagnosis of AIH. Moreover, this study reinforces the close association of extrahepatic autoimmunity among AIH patients and their family. Interestingly, in this study, AIH patients with CEHAID were found to have a lower prevalence of SMA reactivity, less ascites and coagulopathy and milder degree of fibrosis on histology at diagnosis. Although these do not seem to have notable impact on the clinical outcomes of AIH, further evaluation may be warranted to delineate the intricate interplay between extrahepatic autoimmunity and the heterogeneity of clinical phenotype in AIH.

References

1. Gleeson D, Heneghan MA, British Society of Gastroenterology. British Society of Gastroenterology (BSG) guidelines for management of autoimmune hepatitis. Gut. 2011;60(12):1611–29.

2. Heneghan MA, Yeoman AD, Verma S, Smith AD, Longhi MS. Autoimmune hepatitis. Lancet. 2013;382(9902):1433–44.

3. European Association for the Study of the Liver. EASL Clinical Practice Guidelines: Autoimmune hepatitis. J Hepatol. 2015;63(4):971–1004.

4. Johnson PJ, McFarlane IG. Meeting report: International Autoimmune Hepatitis Group. Hepatol. 1993;18(4):998–1005.

5. Alvarez F, Berg PA, Bianchi FB, et al. International Autoimmune Hepatitis Group Report: review of criteria for diagnosis of autoimmune hepatitis. J Hepatol. 1999;31(5):929–38.

6. Csak T, Folhoffer A, Horvath A, et al. Holmes-Adie syndrome, autoimmune hepatitis and celiac disease: a case report. World J Gastroenterol. 2006;12(9):1485–7.

7. Luth S, Birklein F, Schramm C, et al. Multiplex neuritis in a patient with autoimmune hepatitis: a case report. World J Gastroenterol. 2006;12(33):5396–8.

8. Nobili V, Liaskos C, Luigi G, Guidi R, Francalanci P, Marcellini M. Autoimmune thyroiditis associated with autoimmune hepatitis. Thyroid. 2005;15(10):1193–5.

9. Romanelli RG, La Villa G, Almerigogna F, et al. Uveitis in autoimmune hepatitis: a case report. World J Gastroenterol. 2006;12(10):1637–40.

10. Takahashi K, Takasaki S, Morita C, et al. Autoimmune hepatitis with membranous glomerulonephritis. J Gastroenterol Hepatol. 2001;16(3):356–9.

11. Tojo J, Ohira H, Abe K, et al. Autoimmune hepatitis accompanied by systemic lupus erythematosus. Intern Med Tokyo Jpn. 2004;43(3):258–62.

12. Yamaike N, Saigo K, Imoto S, et al. Autoimmune hepatitis accompanied by idiopathic thrombocytopenic purpura and Sjögren's syndrome. Intern Med Tokyo Jpn. 2002;41(1):72.

13. Bittencourt PL, Farias AQ, Porta G, et al. Frequency of concurrent autoimmune disorders in patients with autoimmune hepatitis: effect of age, gender, and genetic background. J Clin Gastroenterol. 2008;42(3):300–5.

14. Muratori P, Fabbri A, Lalanne C, Lenzi M, Muratori L. Autoimmune liver disease and concomitant extrahepatic autoimmune disease. Eur J Gastroenterol Hepatol. 2015;27(10):1175–9.

15. Teufel A, Weinmann A, Kahaly GJ, et al. Concurrent autoimmune diseases in patients with autoimmune hepatitis. J Clin Gastroenterol. 2010;44(3):208–13.

16. van Gerven NMF, Verwer BJ, Witte BI, et al. Epidemiology and clinical characteristics of autoimmune hepatitis in the Netherlands. Scand J Gastroenterol. 2014;49(10):1245–54.

17. Gregorio GV, Portmann B, Reid F, et al. Autoimmune hepatitis in childhood: a 20-year experience. Hepatol. 1997;25(3):541–7.

18. Al-Chalabi T, Underhill JA, Portmann BC, McFarlane IG, Heneghan MA. Impact of gender on the long-term outcome and survival of patients with autoimmune hepatitis. J Hepatol. 2008;48(1):140–7.

19. Al-Chalabi T, Portmann BC, Bernal W, McFarlane IG, Heneghan MA. Autoimmune hepatitis overlap syndromes: an evaluation of treatment response, long-term outcome and survival. Aliment Pharmacol Ther. 2008;28(2):209–20.

20. Yeong TT, Lim KHJ, Goubet S, Parnell N, Verma S. Natural history and outcomes in drug-induced autoimmune hepatitis. Hepatol Res. 2016; 46(3):79-88.

21. Batts KP, Ludwig J. Chronic hepatitis. An update on terminology and reporting. Am J Surg Pathol. 1995;19(12):1409–17.

22. Johnson PJ, McFarlane IG, Williams R. Azathioprine for long-term maintenance of remission in autoimmune hepatitis. N Engl J Med. 1995;333(15):958–63.

23. Homberg JC, Abuaf N, Bernard O, et al. Chronic active hepatitis associated with antiliver/kidney microsome antibody type 1: a second type of "autoimmune" hepatitis. Hepatol. 1987;7(6):1333–9.

24. van Gerven NM, Bakker SF, de Boer YS, et al. Seroprevalence of celiac disease in patients with autoimmune hepatitis. Eur J Gastroenterol Hepatol. 2014;26(10):1104–7.

25. Watt FE, James OFW, Jones DEJ. Patterns of autoimmunity in primary biliary cirrhosis patients and their families: a population-based cohort study. QJM. 2004;97(7):397–406.

26. Couto CA, Bittencourt PL, Porta G, et al. Antismooth muscle and antiactin antibodies are indirect markers of histological and biochemical activity of autoimmune hepatitis. Hepatol. 2014;59(2):592–600.

27. Golding PL, Smith M, Williams R. Multisystem involvement in chronic liver disease. Studies on the incidence and pathogenesis. Am J Med. 1973;55(6):772–82.

28. Al-Chalabi T, Boccato S, Portmann BC, McFarlane IG, Heneghan MA. Autoimmune hepatitis (AIH) in the elderly: a systematic retrospective analysis of a large group of consecutive patients with definite AIH followed at a tertiary referral centre. J Hepatol. 2006;45(4):575–83.

29. Czaja AJ, Donaldson PT. Gender effects and synergisms with histocompatibility leukocyte antigens in type 1 autoimmune hepatitis. Am J Gastroenterol. 2002;97(8):2051–7.

30. Abe M, Mashiba T, Zeniya M, et al. Present status of autoimmune hepatitis in Japan: a nationwide survey. J Gastroenterol. 2011;46(9):1136–41.

31. Tanriverdi F, Silveira LFG, MacColl GS, Bouloux PMG. The hypothalamic-pituitary-gonadal axis: immune function and autoimmunity. J Endocrinol. 2003;176(3):293–304.

32. Prahalad S, Shear ES, Thompson SD, Giannini EH, Glass DN. Increased prevalence of familial autoimmunity in simplex and multiplex families with juvenile rheumatoid arthritis. Arthritis Rheum. 2002;46(7):1851–6.

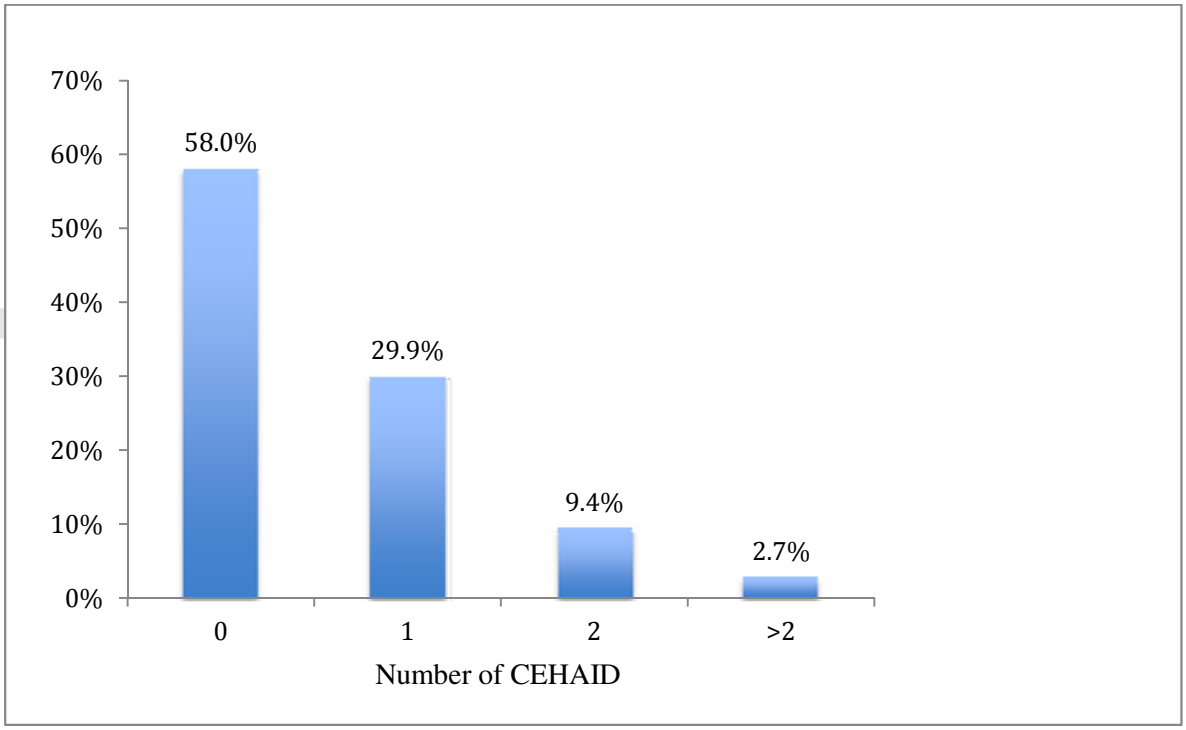

Fig. 1: Distribution of AIH patients with no, one, two and more additional concurrent extrahepatic autoimmune diseases (CEHAIDs)

Table 1. Frequency of CEHAID in AIH patients.

CEHAID	Frequency, n=562
Autoimmune thyroid disease	101 (18%)
Hypothyroidism*	79 (14.1%)
Hyperthyroidism†	22 (3.9%)
Type 1 diabetes mellitus	21 (3.7%)
Connective tissue disorders	69 (12.3%)
Rheumatoid arthritis	19 (3.4%)
Systemic lupus erythematosus	16 (2.8%)
Sjogren's syndrome	16 (2.8%)
Serongative polyarthropathy	8 (1.4%)
Autoimmune skin disease	45 (8%)
Vitiligo	6 (1%)
Alopecia areata	6 (1%)
Gastrointestinal disorders	33 (5.9%)
Ulcerative colitis	21 (3.7%)
Coeliac disease	8 (1.4%)
Hematological disorders	14 (2.5%)
Idiopathic thrombocytopenic purpura	7 (1.2%)
Lung disorders	8 (1.4%)
Pulmonary fibrosis	3 (0.5%)
Neurological diseases	5 (0.9%)
Multiple sclerosis	2 (0.4%)
Renal disease	4 (0.5%)
lupus nephritis	1 (0.2%)
Miscellaneous	
Raynaund's phenomenon	9 (1.6%)

* Hashimoto's thyroiditis (78 patients) and myxedema (1 patient).

† Grave's disease (19 patients) and toxic multinodular goitre (3 patients).

Table 2. Patterns and frequency of CEHAID in AIH patients based on the serological types¶

CEHAID	AIH-1 (n=459) (%)	AIH-2 (n=32) (%)	Autoantibody negative AIH (n=56) (%)	*P*-value
AITD	73 (15.9)	10 (31.3)	18 (32.1)	**0.025***/**0.003**†/NS‡
Hypothyrodism	59 (12.9)	6 (18.8)	14 (25)	**0.014**†/NS*,‡
Hyperthyroidism	14 (3.1)	4 (12.5)	4 (7.1)	**0.024***/NS†,‡
Type1 DM	17 (3.7)	1 (3.1)	3 (5.4)	NS*,†,‡
IBD	23 (5)	0	1 (1.8)	NS*,†,‡
Coeliac disease	6 (1.3)	1 (3.1)	1 (1.8)	NS*,†,‡
SJS	15 (3.3)	1 (3.1)	0	NS*,†,‡
RA	18 (3.9)	0	1 (1.8)	NS*,†,‡
SLE	15 (3.3)	1 (3.1)	0	NS*,†,‡
Skin disease	32 (7)	7 (21.9)	4 (7.1)	**0.009***/NS†,‡

AIH, autoimmune hepatitis; AITD, autoimmune thyroid disease; CEHAID, concurrent extrahepatic autoimmune disease; DM, diabetes mellitus; IBD, inflammatory bowel disease; RA, rheumatoid arthritis; SJS, Sjogren's syndrome; SLE, systemic lupus erythematosus

NS, not significant ($p>0.05$); Values in bold are significant ($p<0.05$).

* *P*-value for AIH-1 vs. AIH-2.

† *P*-value for AIH-1 vs. autoantibody negative AIH.

‡ *P*-value for AIH-2 vs. autoantibody negative AIH.

¶ 15 patients with missing data for autoantibodies were excluded for analysis.

Table 3. Comparison of demographics, mode of onset and presenting signs and symptoms between AIH patients with and without CEHAID

	AIH with CEHAID (n=236)	AIH without CEHAID (n=326)	*P*-value
Age at diagnosis (years)	46 (2-85)	42 (5-82)	0.200
Female, n (%)	201(85.2)	248(76.1)	**0.008**
Caucasian, n (%)	203 (86)	270 (82.8)	0.306
African-carribean, n (%)	21(8.9)	26 (8)	0.679
Asian, n (%)	8 (3.4%)	18 (5.5%)	0.235
Follow-up duration (months)	120 (1-544)	124 (0.25-532)	0.948
Time to diagnosis (months)	3 (0.25-180)	3 (0.25-216)	0.832
Post treatment IAIHG score	22 (11-28)	20 (11-28)	**<0.001**
Definite/Probable AIH	203/33	252/74	**0.009**
AIH-1/AIH-2/ Autoantibody negative	190/17/26	269/15/30	0.357
Mode of onset			
Acute	132/221(59.7%)	182/294 (61.9%)	0.616
Insidious	50/221(22.6%)	60/294 (20.4%)	0.543
Asymptomatic	39/221(17.6%)	52/294 (17.6%)	0.991
Cirrhosis at diagnosis*	72/232 (31%)	115/323 (35.6%)	0.261
Child-pugh score, median (range)	7 (5-11)	7 (5-13)	0.177
Signs and symptoms at index presentation			
Malaise/lethargy	90/221 (40.7%)	128/295 (43.4%)	0.544
Dark urine/pale stool	60/221 (27.1%)	96/295 (32.5%)	0.187
Joint pain	18/221 (8.1%)	9/295 (3.1%)	**0.010**
Pruritus	26/221 (11.8%)	43/295 (14.6%)	0.353
Rash	15/221 (6.8%)	11/295 (3.7%)	0.116
Abdominal pain	57/221 (25.8%)	68/295 (23.1%)	0.472
Jaundice	127/221 (57.5%)	181/295 (61.4%)	0.373
Ascites	14/221 (6.3%)	40/295 (13.6%)	**0.008**
Hepatic encephalopathy	4/221(1.8%)	6/295 (2%)	0.855
Hematemesis	5/221 (2.3%)	9/295 (3.1%)	0.585

AIH, autoimmune hepatitis; GI, gastrointestinal; IAIHG, International Autoimmune Hepatitis Group.

*Based on histology and/or imaging studies

Values in bold are significant (p<0.05).

Table 4. Comparison of biochemical, immunological, hematological parameters and histology at diagnosis between AIH patients with and without CEHAID

	AIH with CEHAID (n=236)	AIH without CEHAID (n=326)	*P*-value
AST, (IU/L) (NR<50)	591(23-4603)	616 (19-3482)	0.605
ALP (IU/L) (NR<130)	196 (67-3588)	184 (23-1677)	0.286
ALP/AST ratio	0.374 (0.03-9.96)	0.371 (0.03-6.80)	0.063
Bilirubin (mmol/L)(NR 5-17)	59 (5-1208)	73.5 (5-1096)	0.196
GGT (IU/L) (NR5-50)	150 (27-3410)	171 (8-1131)	0.213
Albumin (g/L) (NR 35-42)	35 (19-55)	34 (18-48)	0.106
Globulin (g/L)	44.5 (17-96)	46 (14-105)	0.133
Peak IgG (g/L) (NR<18)	24.9 (4.6-70.7)	26.5 (4-89.5)	0.561
INR (NR 0.8-1.2)	1.18 (0.80-3.13)	1.27 (0.87-2.62)	**0.013**
Creatinine (µmol/L) (NR 53-106)	74 (43-210)	72 (38-176)	0.805
Sodium (mmol/L) (NR136-145)	137 (126-143)	137 (80-144)	0.598
Peak ANA	80 (0-10240)	40 (0-5120)	**<0.001**
ANA positivity	163/237 (68.8%)	209/325 (64.3%)	0.519
SMA positivity	111/223 (49.8%)	203/312 (65.1%)	**<0.001**
Peak SMA	0 (0-2560)	40 (0-10240)	0.128
SMA titre <40	120/223 (53.8%)	130/312 (41.8%)	**0.006**
SMA titre >80	67/223 (30%)	128/312 (41.2%)	**0.009**
LKM positivity	17/233 (7.3%)	16/316 (5.1%)	0.197
Histology (Severity of inflammation)*			
Grade <3	31/197 (15.7%)	40/274 (14.6%)	0.734
Grade 3	60 /197 (30.5%)	80/274 (29.2%)	0.768
Grade 4	106/197 (53.8%)	154/274 (56.2%)	0.606
Histology (fibrosis score)†			
Stage 0	5/134 (3.7%)	9/200 (4.5%)	0.731
Stage 1	28/134 (20.9%)	13/200 (6.5%)	**<0.001**
Stage 2	20/134 (14.9%)	20/200 (10%)	0.174
Stage 3	38/134 (28.4%)	82/200 (41%)	**0.018**
Stage 4	43/134 (32.1%)	76/200 (38%)	0.269
Stage 3 + 4	81/134(60.4%)	158/200 (79%)	**<0.001**

ALP, alkaline phosphatase; ANA, antinuclear antibody; AST, aspartate aminotransferase; GGT, gamma glutamyl transferase; INR, international normalized ratio; LKM, liver-kidney-microsomal antibody; NR, normal range; SMA, smooth muscle antibody.

Values in bold are significant (p<0.05).

*91 patients with missing data were excluded from analysis.

†228 patients with either missing data or uncertain fibrosis stage due to severe multi-acinar or panacinar collapse, were excluded from analysis.

Table 5. Comparison of treatment, response to therapy and clinical outcomes in AIH patients with and without CEHAID

	AIH with CEHAID (n=236)	AIH without CEHAID (n=326)	*P*-value
Initial therapy			
Prednisolone alone	190/233 (81.5%)	243/320 (75.9%)	0.114
Prednisolone + Azathioprine	37/233 (15.9%)	63/320 (19.7%)	0.251
Others*	6/233 (2.6%)	14/320 (4.4%)	0.263
Maintenance therapy			
Prednisolone alone	37/224 (16.5%)	39/311 (12.5%)	0.194
Prednisolone + Azathioprine	70/224 (31.3%)	107/311 (34.4%)	0.444
Azathioprine	66/224 (29.5%)	111/311 (35.7%)	0.131
Others†	29/224 (12.9%)	27/311 (8.7%)	0.112
Off therapy	22/224 (9.8%)	27/311 (8.7%)	0.652
Response to treatment and relapses			
Complete response	216/231 (93.5%)	299/316 (94.6%)	0.584
Partial response	10/231 (4.3%)	9/316 (2.8%)	0.350
No response	5/231 (2.2%)	8/316 (2.5%)	0.781
One relapse	39/212 (18.4%)	60/300 (20%)	0.651
≥2 relapses	46/212 (21.7%)	76/300 (25.6%)	0.342
Clinical outcomes[¶]			
Disease progression	69/194 (35.6%)	91/271 (33.6%)	0.656
HCC	7/194 (3.6%)	13/263 (4.9%)	0.491
Total LTX/death	43/194 (22.2%)	71/263(27%)	0.238
Liver-related death[‡]/LT	28/39 (71.8%)	40/69 (58.0%)	0.153

HCC, hepatocellular carcinoma; LT, liver transplantation.

*Second-line therapy such as tacrolimus, mycophenolate mofetil or cyclosphosphamide.
†Different combinations of prednisolone, tacrolimus, mycophenolate mofetil and azathioprine.

[‡]Etiology of death for 6 patients were uncertain.
[¶]105 patients who had lost to follow-up were excluded from analysis.

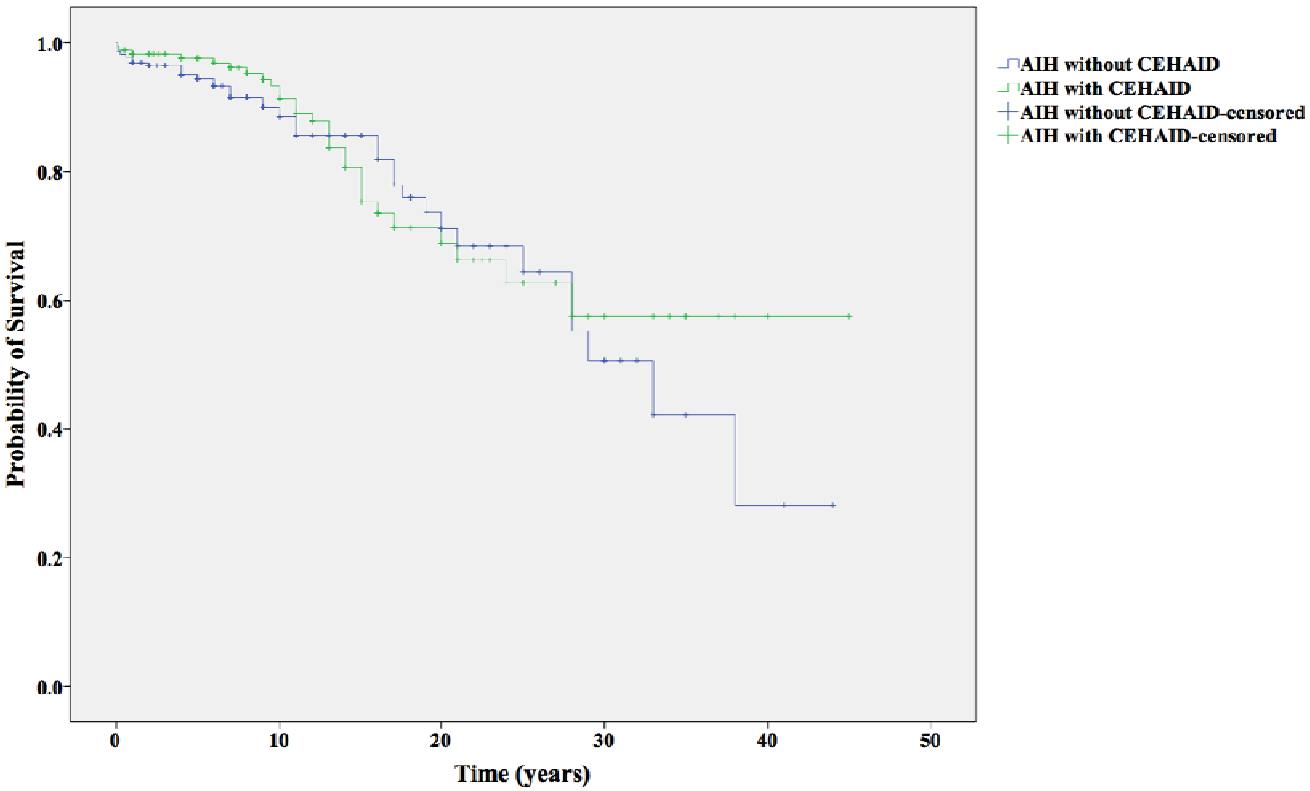

Figure 2. Survival curve of AIH patients with and without CEHAID (P=0.563, log rank)

Table 6. Univariate and multivariate Cox Regression Analysis of Factors associated with reduced survival in AIH patients

Variable*	**Univariate Analysis**		**Multivariate Analysis**	
	Hazard (range)	***P-value***	***Hazard (range)***	***P-value***
Age	1.02 (1.0-1.03)	**0.022**	1.04 (0.99-1.07)	0.078
Encephalopathy	10.02 (3-33.4)	**<0.001**	2.82 (0.32- 24.98)	0.346
Ascites	2.98 (1.52-5.5)	**0.001**	0.85 (0.23-3.15)	0.806
Hematemesis	4.74 (2.09-10.70)	**<0.001**	2.17 (0.25-18.84)	0.482
AST	0.99 (0.99-1.00)	**0.001**	1.00 (0.99-1.00)	0.723
ALP/AST ratio	1.36 (1.19-1.55)	**<0.001**	1.27 (0.95-1.70)	0.105
Albumin	0.93 (0.89-0.97)	**<0.001**	0.88 (0.78- 0.99)	**0.046**
INR	2.24 (1.19-4.22)	**0.013**	2.04 (0.46- 9.14)	0.351
Creatinine	1.02 (1.00-1.04)	**0.013**	1.01 (0.98-1.04)	0.547
Cirrhosis	3.36 (2.00-5.62)	**<0.001**	2.46 (0.71- 8.50)	0.153

*At diagnosis/index presentation